Seasonal variation in dust mite and grass-pollen allergens in dust from the houses of patients with asthma

Thomas A. E. Platts-Mills, M.D., Mary L. Hayden, R.N., Martin D. Chapman, Ph.D., and Susan R. Wilkins, B.Sc. *Charlottesville, Va.*

In order to study seasonal variation in dust-mite allergen, we obtained dust samples from bedding, carpet, and/or sofas in 12 houses in central Virginia, monthly, for 1 year. The houses included those of nine patients with asthma of whom six were allergic to dust mites. Dust samples were assayed with an inhibition radioimmunoassay for mite allergen that detects cross-reacting determinants on Der f *and* Der p *I from* Dermatophagoides farinae *and* D. pteronyssinus, *respectively. The results are expressed as micrograms of antigen P_1 equivalent (AgP_1Eq). The results demonstrate that large seasonal variations in allergen, i.e., more than twentyfold, can occur in dust from all sites and are not restricted to the houses of allergic patients. However, dust from some sites, particularly sofas, remained "high" (>10 μg AgP_1Eq per gram), whereas dust from other sites remained "low" (<1 μg AgP_1Eq per gram) throughout the year. Levels of mite allergen generally started to rise in July about 1 month after the rise in humidity. In August to December, the mean levels of AgP_1Eq in house dust were highly significantly increased relative to April to May. In keeping with this finding, in 31 of 37 sites, the highest level for the year was observed in August through December. In four sites, mite bodies were counted, and the numbers increased sharply in June to July; however, they decreased in September in parallel with falls in humidity but several months before the fall in mite allergen. Ryegrass-pollen allergen in 12 sites was also assayed in house dust, and pollen-allergen levels demonstrated a sharp increase in May or June that fell back to preseason values within 2 months. Dust was also obtained from the houses of 50 patients with acute or severe asthma. The results on these samples suggest that mite-allergic patients are more likely to have attacks in the fall at a time when their houses have >10 μg AgP_1Eq per gram of dust. The magnitude of changes observed seasonally within individual houses and of differences between houses within a close geographic area suggests that interpretation of the relationship between allergic symptoms and mite-allergen exposure will require measurement of mite-allergen levels in individual houses. (J ALLERGY CLIN IMMUNOL 1987;79:781-91.)*

Immediate hypersensitivity to house dust allergens is common among patients with rhinitis, asthma, and atopic dermatitis.[1, 2] Most of these cases are regarded as perennial because they do not exhibit the strict seasonal exacerbations typical of pollinosis. Nonetheless, many practitioners are aware that symptoms related to house dust sensitivity are more likely to exacerbate at certain times of the year. When pyroglyphid mites were first demonstrated to be a major source of house dust allergens, it was reported from The Netherlands that the seasonal increase in mite numbers correlated both with increased humidity and increased symptoms.[3, 4] A similar relationship be-

Abbreviations used

AgP_1Eq:	Antigen P_1 equivalent
BSA:	Bovine serum albumin
Der p I:	Antigen P_1 of *Dermatophagoides pteronyssinus* (Antigen P_1 equal Dp 42 and Dpt 12)
Der f I:	Antigen F_1 of *D. farinae* (Antigen F_1 equal Df 11)
Grass allergen:	Grass-pollen allergen
Lol p I:	Group I protein of ryegrass *(Lolium perene)* pollen (Rye I)
PBS:	Phosphate-buffered saline, pH 7.2
RIA:	Radioimmunoassay

From the Division of Allergy and Clinical Immunology, Department of Medicine, University of Virginia, Charlottesville, Va.

Supported by National Institutes of Health Grant No. A120565, and supported in part by North Atlantic Treaty Organization research grant.

Received for publication June 16, 1986.

Accepted for publication Nov. 7, 1986.

Reprint requests: Thomas A. E. Platts-Mills, M.D., University of Virginia, Division of Allergy and Clinical Immunology, Medical Center, Box 225, Charlottesville, VA 22908.

tween humidity, mites, and asthma symptoms was reported from East Africa,[5] but little seasonal variation was apparent in England.[6] Several studies have demonstrated a seasonal rise in mite numbers in North America that correlated well with humidity[7-9]; however, the peak incidence of mites in July and August did not correlate closely with increased respiratory symptoms in mite-allergic individuals that occurred August to January.[8, 9] Of the many different allergens that can contribute to house dust, only a few would be expected to be seasonal, eg., mites, pollen, and fungi.

Exposure to mite allergens can be assessed by counting the numbers of mite bodies in house dust samples.[9, 10] There are two problems with this. First, the procedure requires microscopic identification and is consequently slow. Second, the number of mite bodies is only indirectly related to the total mite-allergen level because much of the mite allergen is fecal.[11, 12] Alternatively, dust-mite allergens can be measured in house dust with either RAST inhibition or specific immunoassays for purified mite allergens.[12-15] The two most common dust mites in North America are *Dermatophagoides pteronyssinus* and *D. farinae*.[7-10, 16, 17] Of the allergens produced by these mites, only *Der p* I and *Der f* I have been fully purified and can be measured in absolute units.[18-20] We recently reported an inhibition RIA for measuring both *Der p* I and *Der f* I in mite and house dust extracts.[21] This assay detects cross-reacting determinants on each allergen that are collectively referred to as AgP_1Eq. The results for AgP_1Eq correlate well with other assays of the mite-allergen content of house dust or mite counts.[21, 22] In the present study, we assayed AgP_1Eq in dust samples collected from 12 houses monthly for 1 year and from the houses of 54 additional patients with asthma. The results demonstrate a highly significant increase in mite allergen from July to December. In contrast, the peak levels of ryegrass-pollen allergen (grass allergen) in house dust were found during May to July.

METHODS

Patients and house visiting

Houses visited monthly included those of seven patients with asthma who were allergic to dust mites. Mite allergy was dominant in four of these patients: two patients who had asthma but were skin test and RAST negative to dust mites and three asymptomatic nonallergic control subjects. Five houses were in Charlottesville, Va., and seven houses were in the surrounding area. All but one house were within 10 miles of Charlottesville. Visits were carried out by one of us, or in four cases, the patients were trained to collect their own dust samples. Samples were collected with a hand-held vacuum cleaner Douglas Hand Vac AL6701 (Douglas Products, Walnut Ridge, Ark.) with a filter holder behind the cleaner head (MS filter, type 681, Cambridge, U. K.).[21, 23] Samples were collected onto tissues from an area of 1 m^2 for 1 minute and stored at 4° C. In most cases three samples were collected from each house: bedding, bedroom floor, and family room floor. In addition, serial samples were obtained from eight sofas and three upholstered chairs (referred to in the text as sofas). In houses in which no carpeting was available, more sofas were studied. For 11/12 houses, temperature and relative humidity was recorded at each visit with a hand-held humidity meter. The meters were calibrated, and results in houses were checked with a psychrometer (Bendix Model No. 566, Bendix Environmental and Process Instr., Lewisburg, W. Va.). Outside absolute humidity, relative humidity, and temperature for 1984 and 1985 were obtained both from Charlottesville airport and also for Roanoke, Va., from the National Oceanic and Atmosphere Administration, National Climatic Data Center, Asheville, N. C.

In addition, 81 visits were made to the homes of patients with asthma or rhinitis who presented to the adult allergy and pulmonary clinics of the University of Virginia. Fifty-four visits (involving 48 patients and including three visits that were part of the year-round study) occurred at a time when the patient either had severe chronic asthma requiring systemic steroids or within 2 weeks of an exacerbation requiring increased treatment for asthma. In each of these cases, asthma was documented by history, physical signs, a peak expiratory flow rate <60% of predicted, and 20% increase after bronchodilator therapy. Dust was also obtained from the houses of 23 patients with a history of perennial rhinitis. All patients had been skin tested with the prick technique with a range of inhalant allergens including *D. farinae*, cat dander, *Alternaria*, *Aspergillus*, *Cladosporium*, and mixed cockroach extract from Hollister-Stier (Spokane, Wash.), and with mixed grass pollen and ragweed pollen from Greer Laboratories (Lenoir, N. C.). Skin tests were graded by wheal size, and wheals with a mean diameter of 3 mm greater than wheals of control subjects were regarded as positive. Dust-mite sensitivity was regarded as dominant when the prick test response to *D. farinae* was at least 5 by 5 mm, and the mean diameter was at least 2 mm greater than the response to any other allergen.

Extraction of dust and mite counts

The techniques used for handling and extracting dust samples as well as those for counting mites have been reported in full previously.[10, 12, 21] Briefly, samples were sieved through an 0.3 mm mesh screen to obtain fine dust. Mites were recovered from a 100 mg portion and counted microscopically.[24] Dust from nine of the houses was examined for mites, and their species were identified on at least one occasion. In eight cases the dust contained both *D. farinae* and *D. pteronyssinus*. The dust in three of these houses also contained Cheletidae, and one house contained small numbers of Tarsonimidae. One house contained *D. farinae* only. A separate 100 mg sample was extracted with 2 ml of borate-

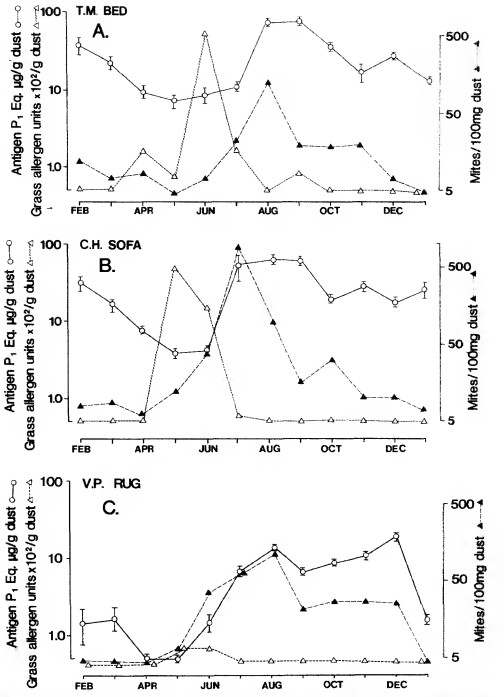

FIG. 1. Results of mite allergen, mite bodies, and grass-pollen allergen in dust samples from three individual sites in three houses **A, B,** and **C** are presented. Mite allergen ($Ag P_1 Eq$) was measured by RIA (o——o), and the range of results observed in two assays is indicated by the *bars*. Mite bodies per gram of dust were counted by microscopy; >90% of identified mites were *Dermatophagoides* species and <10% were alive (▲ - - - ▲). Grass-pollen allergen was measured by a modified direct RAST. We estimate that the units represent ~0.1 ng of Rye I (△ - - - △).

buffered saline pH 8.0, by rotation for 2 hours. After centrifugation, the supernatant was stored at −20° C. Samples with smaller total weight were extracted in a proportional volume of saline. Samples with <50 mg were extracted with 1.0 ml, and results were adjusted accordingly. Samples with <10 mg of fine dust were excluded from analysis, and sites were only included where at least nine monthly samples provided an adequate quantity of fine dust. Of 43 sites

TABLE I. Monthly measurements of ryegrass pollen allergen in 12 sites (units per gram of dust)*

House	Site	Assay	Mar	Apr	May	June	Jul	Aug	Sept	Oct
T. M.	Carpet	i	<	<	<	4660	310	<	100	<
		ii	<	<	<	6500	105	<	<	<
T. M.	Bed	i	<	168	76	24,000	2900	<	<	<
S. W.	Carpet	i	<	<	<	310	<	<	<	<
		ii	<	<	<	452	<	<	<	<
C. H.	Sofa	i	<	<	5100	2000	64	<	<	<
		ii	<	<	5200	1600	<	<	<	<
C. H.	Bed	i	<	<	690	470	245	<	<	<
C. M.	Curtains	i	<	<	6020	<	<	<	<	<
V. P.	Rug	i	<	<	68	68	<	<	<	<
V. P.	Bed	i	<	<	380	80	<	<	<	<
L. C.	Chair	i	<	<	<	3000	720	116	140	<
		ii	<	<	<	4500	540	100	80	<
L. C.	Sofa	i	<	<	<	8200	120	60	<	<
D. R.	Sofa	i	<	<	1740	1640	470	52	<	<
V. L.	Sofa	i	<	<	380	1820	1960	<	<	<
Geometric mean†			51	57	275	977	208	53	56	<50

*Samples were assayed with a modified direct RAST with a capturing antibody on the solid phase. All samples were assayed at at least two dilutions, and the year-round samples at each site were assayed in a single assay. Samples from four sites were assayed on two separate occasions (i) and (ii), and the results are presented to illustrate the variability of the assay.

†For calculation of geometric mean, the results for the sites that were assayed twice were meaned first. The standard error of the geometric mean values is presented in Fig. 4.

studied, six were excluded because of an inadequate number and/or quantity of samples.

RIA for AgP_1Eq

The RIA for mite allergen has been described in full elsewhere.[21] In brief, the assay identifies cross-reacting determinants on *Der p* I of *D. pteronyssinus* and *Der f* I of *D. farinae*, and the results are expressed in micrograms of AgP_1Eq per gram of fine dust. Each sample was assayed at two dilutions, 1:6 and 1:36, and when it was necessary, samples were repeated at higher or lower dilutions. The minimum sensitivity of the assay is ~10 ng/ml, and since 100 mg of dust is extracted in 2.0 ml, the minimum sensitivity is ~0.2 μg/gm of fine dust. The inhibition RIA uses rabbit antibodies to *Der f* I specifically purified on a *Der p* I column, i.e., directed against cross-reacting determinants. A control curve was carried out in parallel with each assay with a *D. pteronyssinus* extract substandardized from the World Health Organization International Standard (NIBSC 82/518) that we have previously assayed and contains 12 μg AgP_1Eq per ampule.[25,26] This assay has an intra-assay variability of ±15% and an interassay variability of ±17%.[21]

RIA for grass allergen in house dust

The assay used was a modified direct RAST with a solid-phase antibody for capturing allergen. In the first phase, a hyperimmune rabbit antiserum to ryegrass pollen was applied to microtiter plates (Dynatech, Alexandria, Va.). One hundred microliters of a 1:3000 dilution of the antiserum diluted in a carbonate bicarbonate buffer, pH 9.0, was applied to each well and incubated overnight. After washing, the wells were "blocked" with 3% BSA in Tween 20 and PBS, pH 7.2, for 1 hour. The samples of dust extract or standard diluted in 3% BSA in PBS and Tween 20 were added in 100 μl and incubated for 6 hours at room temperature. After washing three times, 100 μl of a 1:10 dilution of a serum known to contain a very high titer of IgE antibody to grass pollen was added and incubated overnight at room temperature. After washing four times, 100 μl of radiolabeled goat anti-IgE (containing ~7 ng of anti-IgE) diluted in 50% fetal calf serum in PBS with 0.05% Tween 20 was added for an additional 6 hours. Finally, plates were washed five times, and the wells were counted in a Micromedic 4 channel gamma counter (Micromedic Systems, Inc., Horsham, Pa.). In general, dust samples were assayed diluted 1:2 and 1:10, and standard ryegrass extract (Greer Laboratories) was assayed in parallel with serial twofold dilutions from 1:2000 to 1:16,000,000. This extract was arbitrarily allotted 10 million units of ryegrass allergen. Values for dust extracts were interpolated from this control curve, and results were expressed in units per gram of dust. Experiments with purified *Lol p* I (equal to group I protein of ryegrass pollen; kindly provided by Dr. David Marsh) suggest that the unit is equivalent to ~0.1 ng of this major grass allergen.

Statistics

The levels of allergen at different sites and at different times of the year were compared by Student's unpaired

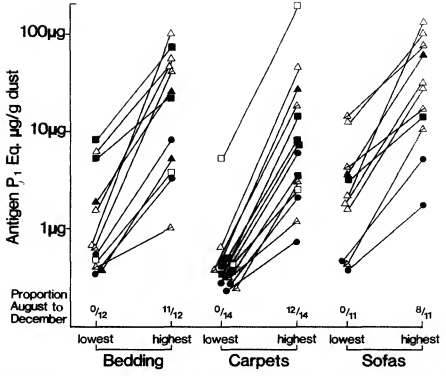

FIG. 2. Maximum and minimum concentrations of AgP_1Eq/gm of dust recorded for 37 separate sites are presented. *Triangles* indicate the house dust mite–allergic patients with asthma, *squares* indicate nonallergic control subjects, and *circles* indicate the two nonallergic patient with asthma. The *numbers* at the bottom of the figure indicate the proportion of sites where the lowest (or highest) value was observed during August through December. Also indicated are the air-conditioning used in the houses. *Solid* symbols indicate houses with central air-conditioning, ●, ▲, ■, *open symbols* indicate houses with no air-conditioning (△, □), whereas the houses with room air-conditioning only are indicated by *slashed symbols* (⧋).

t test on the log values, and ranges were presented as the standard error of the log mean. The incidence of values >10 µg and >2 µg was compared with chi-squared test.

RESULTS

All dust samples were sieved and assayed for AgP_1Eq per gram of fine dust. In addition, mite numbers were counted in samples from four sites in four houses, and ryegrass-pollen allergen was assayed in 12 sites from seven houses. The full results for three individual sites demonstrate a marked increase in mite allergen in July that coincided with a sharp increase in mite numbers (Fig. 1, *A*, *B*, and *C*). At each of these sites, mite numbers increased to >1000 mites per gram of fine dust and subsequently fell faster than the allergen content that remained high until December. Grass allergen rose sharply in May and June, fell in July, and was absent or very low for the rest of the year (Fig. 1 and Table I). Pollen-allergen levels rose by more than tenfold in nine of 12 sites studied (Table I). Adequate serial samples were obtained from 12 beds in 12 houses, from 14 carpets or rugs in nine houses, and from 11 sofas in nine houses. The full results for AgP_1Eq are presented for three sites in each of six houses (Table II). When the lowest and highest values recorded at each site were examined, it was obvious that variation occurred at all sites (Fig. 2). However, the range of results at any one site varied from threefold to more than seventyfold. Also illustrated in Fig. 2 is the incidence of sites where the highest value was in the months of August to December (overall 32/37), whereas none of the lowest values were in these 5 months ($p < 0.001$). The mean values for bedding, carpets, and sofas analyzed in bimonthly periods demonstrated a consistent pattern. Sofas were the highest, bedding was next, and carpets were the lowest (Fig. 3). The difference between sofas and carpets was highly significant ($p < 0.001$) for each bimonthly period except June to July ($p < 0.01$). Furthermore, for carpets, bedding, and sofas, the mean values in October through November were significantly higher than in April through May (Student's

TABLE II. Monthly measurements of dust mite allergen AgP_1 Eq at three sites in six houses*

House	Site	Months of 1984/1985							
		Feb	Mar	Apr	May	Jun	Jul	Aug	Sep
V. P.†									
Asthma,	Bed	12	1.9	0.6	0.9	2.3	2.6	44	36
mite	Carpet	0.7	1.1	0.4	0.4	1.1	5.7	14	6.3
	Sofa	3.9	12	6.3	13	6.2	5.4	14	12
L. C.									
Asthma,	Bed	—	47	2.2	0.9	5.5	0.6	1.9	7.9
mite	Sofa	12	3.7	3.1	1.8	1.7	7.7	8.2	17
	Chair	13	6.6	4.9	2.3	3.8	4.9	8.7	31
D.R.									
Asthma,	Bed	27	55	23	5.9	6.2	11	9.9	8.4
mite	Carpet	4.2	2.9	4.6	0.7	0.5	11	15	18
	Sofa‡	16	14	76	22	46	33	43	82
V. L.									
Asthma,	Bed	0.2	1.0	0.4	0.4	0.4	0.4	0.9	0.5
cat	Carpet‡	0.2	0.2	0.6	0.2	0.5	0.4	0.8	0.5
	Sofa	1.8	0.4	0.5	0.8	2.6	1.0	4.9	3.9
E. W.									
Asthma,	Bed	3.4	1.0	0.9	0.6	0.5	—	8.1	0.7
nonallergic	Carpet	0.6	0.2	0.2	0.5	0.4	—	<0.2	<0.2
	Sofa	0.5	0.4	0.5	0.8	0.4	—	0.6	1.0
T. M.									
Control,	Bed	38	22	9.4	7.1	8.9	11	74	75
nonallergic	Carpet	0.3	1.3	0.6	0.6	1.0	7.0	10	11
	Carpet	0.9	0.4	0.5	<0.2	0.4	0.7	1.7	3.5

*Values are micrograms of antigen P_1 equivalent per gram of sieved dust. Dashes indicate samples that were of inadequate quantity to assay (<10 mg of dust) or were not obtained.

†Patients are indicated with diagnosis and their dominant inhalant sensitivity. Patient V. P. also had atopic dermitis.

‡The mean value for 12 samples from sofa of D. R. was 39 (22 to 69), and the mean value for 12 samples from the carpet of V. L. was 0.32 (0.2 to 0.54) μg AgP_1 Eq/gm (geometric mean with 95% confidence limits of the mean).

t test, $p \leq 0.001$). Also illustrated in Fig. 3 are the mean monthly levels for grass allergen, emphasizing the contrast between the two seasons. In Fig. 2, the houses without air-conditioning are indicated as *open symbols*. Although six of the highest values were recorded in nonair-conditioned houses, high values were also recorded in air-conditioned houses. Interestingly, two of the houses that had no air-conditioning had no carpets, but had sofas. This meant that there was a slightly disproportionate number of sofas from nonair-conditioned houses that may in part explain the higher levels of allergen found in sofas.

Humidity measurements were obtained in each house at the time of visits, and the mean values for indoor humidity are illustrated in Fig. 4. We also obtained daily values from Charlottesville airport and Roanoke for 1984 to 1985. Calculated as mean monthly absolute humidity, the outside values demonstrate greater variation than the indoor values but correlate well overall (Fig. 4). The maximum humidity occurred in June, July, and August and appears to precede the rise in mite allergen by approximately 1 month. Mean temperatures in the 12 houses were 67 ± 3.9° F in February to April, 73 ± 3.5° F in June to August, and 68 ± 4.1° F in October to December.

We have previously suggested that 10 μg AgP_1Eq per gram of dust is a level of mite allergen that is commonly associated with symptoms in mite-allergic patients.[21,23] Significant increases in the percentage of sites with >10 μg/gm or >2 μg/gm of dust were found in September to December (Fig. 5). Among individual houses, the percentage of dust samples with high mite-allergen levels demonstrated dramatic variation. In two houses only 10% and 19% of samples had >2 μg AgP_1Eq per gram of dust, and the levels never exceeded 10 μg/gm at any time during the year. In two other houses, almost all (93% and 97%) samples had >2 μg AgP_1Eq, and 62% and 78% of samples had >10 μg/gm.

Months of 1984/1985			
Oct	Nov	Dec	Jan
22	25	5.3	17
8.4	11	19	1.3
12	13	9.0	7.2
—	6.0	10	2.4
—	25	30	23
—	30	24	13
9.3	45	32	27
11	48	31	5.0
64	71	43	35
1.8	1.0	0.7	1.1
<0.2	0.4	<0.2	<0.2
6.9	10	—	4.2
1.8	0.2	0.5	1.2
2.3	1.1	0.9	0.6
1.0	1.8	1.4	0.6
36	17	28	13
2.5	9.7	5.6	1.4
3.5	3.7	3.6	1.2

During the year of this study, 1984, four of the mite-allergic patients whose houses we studied had an exacerbation of their asthma that required additional treatment. In each case the exacerbation occurred in the 5-month period September to December and at a time when at least one site in their house contained 10 μg AgP_1Eq per gram of dust. However, this relationship is not simple because in two of the cases, the mite-allergen level had been high for 3 months previously, and in another case the patient was also allergic to cat dander. During the period 1983 to 1985, we made 58 additional visits to the houses of patients with asthma to collect dust. Overall, we made 54 house visits at a time when the patient was either chronically wheezing and receiving systemic steroids or within 2 weeks of an acute episode of asthma requiring additional treatment. High levels of dust-mite allergen were very commonly found in the houses of patients with asthma whose dominant skin sensitivity was to dust mites (Fig. 6); in 16/18 visits occurring within 2 weeks of an exacerbation, AgP_1Eq was >10 μg/gm. In house dust from the houses of patients with asthma who were either nonallergic or had multiple positive skin tests, there was a significantly greater incidence of mite-allergen levels <10 μg/gm (17 of 36; chi-squared, $p < 0.01$). However, this difference can in large part be explained by the fact that the house visits of the "nonmite dominant" patients occurred more commonly in January to July (Fig. 6). Indeed, 13 of the 17 houses in which allergen levels were <10 μg/gm were visited in January to July. These results also confirm that high and very high levels of mite allergen are not restricted to the houses of mite-allergic patients.

DISCUSSION

The results presented here demonstrate highly significant increases in the mite-allergen content of house dust during the period August to December. The AgP_1Eq RIA demonstrates no significant cross-reactivity with a large range of other allergens, including animal danders, pollens, insects, storage mites, and fungi.[21] Recently, we found an excellent correlation between measurements of mite-allergen content of house dust by RAST inhibition and AgP_1Eq content (for 21 samples, $r = 0.91$, $p < 0.001$).[22] In our previous studies on *D. pteronyssinus* growth in culture, the total quantity of *Der p* I correlated well with the number of mites regardless of the culture medium.[11] In house dust samples, the correlation between mite numbers and AgP_1Eq content was highly significant but demonstrates considerable scatter, both in the present study and in previous studies.[12, 21-23] The variation in mite numbers may in part reflect the fact that mites avoid dry conditions and will thus move away from the surface of furniture as drying occurs. Live mites will only be present on the surface during periods of high relative humidity. Thus, the number of mites found in surface dust may not closely relate to either the number of mites living within that piece of furniture, bedding, etc. or the total quantity of allergen in the dust. It is important to remember that most of the mite bodies found in house dust are dead and that the allergen in the bodies does not explain >20% of the mite allergen found in house dust.[2, 21]

In the present study, the sharp rise in mite numbers and mite allergen occurred in July and August that is 1 month after the rise in mean outdoor absolute humidity. This delay is in keeping with previous results and presumably reflects the time taken for mites to increase their numbers.[8, 9, 17] The houses were also hotter during the summer, and the increase in June to August, ~6° F, was certainly sufficient to increase mite growth.[4, 10] Overall, it is believed that absolute humidity is the best single guide to excess mite growth.[3, 4, 10, 27-29] The interesting question is whether a particular level of indoor humidity could be defined that should be avoided in order to prevent excessive mite growth. In the present study, the indoor humidity measurements were only made once per month; however, an indoor absolute humidity of 9 gm/kg appears

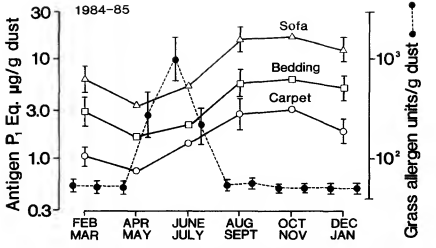

FIG. 3. Geometric mean values are presented for mite allergen in dust from 11 sofas (△——△), 12 beds (□——□), and 14 carpets (○——○). Values were analyzed in bimonthly periods, and the standard error of the geometric mean is presented for three typical bimonthly periods. For each type of sample, the values in October through November were significantly higher than in April through May (two-sided t test, $p < 0.005$). The difference between carpets and sofas was highly significant for each bimonthly period ($p < 0.005$), but the difference between bedding and the other sites did not reach significance. Also presented are the results for grass-pollen allergen, geometric mean ± SEM for 12 sites analyzed on a monthly basis (●- - -●).

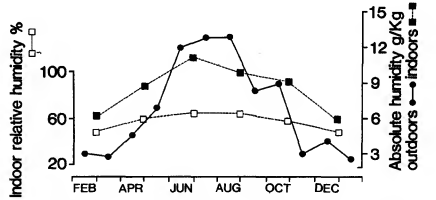

FIG. 4. Mean indoor relative (□- - -□) and absolute humidity (■- - -■) for 11 houses analyzed on a bimonthly basis. These values represent the mean of measurements made on 1 day per month. Outdoor absolute humidity values are mean values from four measurements per day made at Roanoke airport (●——●) (100 miles from Charlottesville, Va., with very similar weather). Values provided by National Climatic Data Center, Asheville, N. C.

to be critical, and this is similar to values reported from Europe.[4, 27] Although mite allergen rose within 1 month of the rise in humidity, the fall in allergen levels was delayed by as much as 3 months. This phenomenon was consistent, but the reasons for it are not immediately apparent. It is possible that mite allergen that accumulated during the period of maximum mite growth takes many months to remove. This appears unlikely because we have previously observed rapid falls (75% in 1 month) in mite allergen recovered from carpets treated with the acaricide pirimiphos methyl that does not have any effect on the allergen itself.[30] At the present time, it appears more likely that live mites persist within furnishings and continue to produce allergen that comes to the surface largely in the form of fecal pellets.[11, 12] Thus, we assume that the reduction in the number of mites reflects drying of surface areas but that major falls in allergen production do not occur until most of the fabric has become dry, which may take several months.[4]

The rapid fall in grass allergen that occurred in July suggests that cleaning is effective at removing pollen that has settled on furniture. This contrasts with a

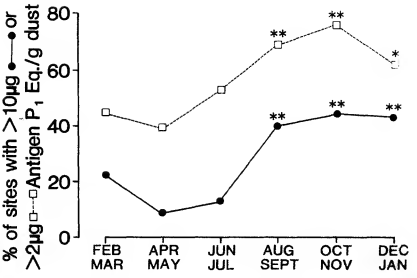

FIG. 5. Percentage of dust samples with >10 μg (●—●) or >2 μg AgP_1Eq/gm (□ - - - □) presented on a bimonthly basis. Values that were significantly higher than the values for April through May are indicated by ** ($p < 0.005$) and * ($p < 0.05$).

recent report that oak-pollen allergen can be detected outside many months after the pollen season.[31] The present studies confirm the presence of pollen allergen in house dust but were not detailed enough to compare grass-allergen levels with the pollen count.[32, 33] We have estimated that 1 unit of grass allergen is equivalent to 0.1 ng of *Lol p* I (equal to Rye I); therefore, 1000 units of grass allergen could represent several hundred pollen grains in a gram of house dust (assuming 0.1 to 0.3 ng of *Lol p* I per pollen grain).[34] In order to study the influence of indoor pollen on respiratory symptoms, it would be necessary to do weekly studies throughout the pollen season. Nonetheless, it appears likely that the level of indoor pollen should be considered as one of the variables in the relationship between pollen counts and respiratory symptoms.[35]

In the present study, two nonallergic individuals remained asymptomatic despite mite-allergen levels of 75 μg/gm and 204 μg/gm, which is in keeping with previous results from London.[12] Our results do not confirm the study from Denmark that suggested that the houses of nonallergic individuals have consistently lower mite numbers than houses of mite-allergic patients with asthma.[27] However, in a cohort study on young children in England, the incidence of IgE and IgG antibody to mites was increased in houses with >5 μg of *Der p* I per gram of dust.[36] The effect of mite allergen on the lungs of allergic patients will be influenced by several other variables, including the quantity that becomes airborne and the duration of exposure.[37] Thus, we would not expect that mite-allergic patients with asthma would present with their worst symptoms as soon as mite-allergen levels rise in house dust. In the present study, dust samples were obtained on 18 occasions from the houses of mite-allergic patients with asthma who were having continuous symptoms or exacerbations. In 16 of these houses, at least one sample contained >10 μg AgP_1Eq per gram. This result suggests that mite-allergic patients have attacks more often at times of the year when there are high levels of mite allergen in their houses, whereas nonmite-allergic patients with asthma are equally likely to have attacks at times of the year when their houses have low levels of mite allergen. However, mite-allergic patients with asthma do not necessarily have acute symptoms when there are high levels of allergen in their houses. Thus, a high level of mite allergen in house dust appears to be but one of several contributing factors in these patients. We have analyzed the results with 10 μg AgP_1Eq per gram as a cutoff, but this level is arbitrary. Allergen levels in homes should perhaps be considered in the same way as pollen counts for pollen-allergic patients. There is certainly no specific level of ragweed pollen at which ragweed-sensitive patients will develop symptoms. We suggest that 10 μg AgP_1Eq per gram of dust is approximately equivalent to a pollen count of 200/m^3. This view is in keeping with previous findings in London that mite-allergic patients improved when they moved from their home (mean allergen level 13.6 μg *Der p* I per gram) to live in an "allergen-free" room (mean allergen level 0.23 μg *Der p* I per gram).[38] Thus, many sites in the present study varied seasonally from a level that was close to that in an allergen-free room to levels that appear to be associated with marked increase in symptoms in mite-allergic patients.

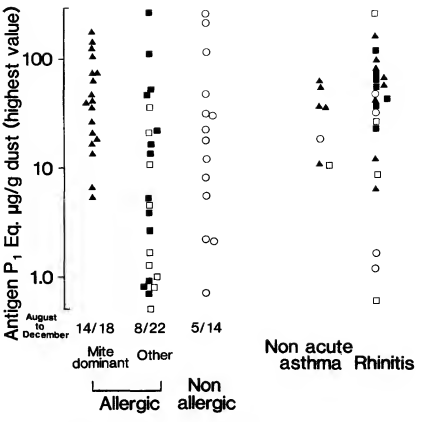

FIG. 6. Dust mite–allergen levels in the houses of patients with asthma sampled within 2 weeks of an exacerbation or when they were chronically wheezing while they were receiving steroids. Results presented are the highest value observed in three samples. Patients were classified according to the results of skin testing with a range of inhalant allergens: patients in whom dust mite was the only skin sensitivity or the strongest sensitivity (▲), patients who gave positive skin tests to dust mites but also gave positive results to other allergens (■), patients who were negative to dust mites but positive to other allergens (□), and patients who were skin test negative to inhalant allergens (○). For the patients with acute or severe asthma, the incidence of visits during August to December is indicated. Also presented are results for seven patients with asthma who were in remission at the time of the visit and for 22 cases of rhinitis.

In conclusion, the mite-allergen content of house dust demonstrates large seasonal variations that follow changes in indoor absolute humidity. On an average, the highest levels of allergen were found in sofas, but high levels and significant seasonal variation was observed in bedding, sofas, and carpets. In only three sites was the seasonal change less than fivefold, and in nine sites there was more than a twentyfold change in mite-allergen content. In 86% of the sites, the highest level was observed in the 5 months from August to December. The magnitude of these changes suggests that accurate interpretation of the relationship between house dust exposure and allergic symptoms will require the use of assays for monitoring both mite and other allergen levels in individual houses.

We are grateful to Dr. David Marsh for providing purified *Lol p* I and antisera to ryegrass allergens, to Dr. Sharon Esau, Dr. Paul Surratt, Dr. John Guerrant, and Dr. Dudley Rochester for allowing us to study patients under their care, to Gail Rose for excellent technical assistance, and to Nancy Malone for preparing the manuscript.

REFERENCES

1. Kern A. Dust sensitization in bronchial asthma. Med Clin North Am 1921;5:751.
2. Tuft LA. Importance of inhalant allergens in atopic dermatitis. J Invest Dermatol 1949;12:211.
3. Voorhorst R, Spieksma FThM, Varekamp H, Leupen MJ, Lyklema AW. The house dust mite (*Dermatophagoides pteronyssinus*) and the allergens it produces: identity with the house dust allergen. J ALLERGY 1967;39:325.
4. Spieksma FThM. The house-dust mite *Dermatophagoides pteronyssinus* (Trouessart, 1897). In: Voorhorst R, Spieksma FThM, Varekamp H, eds. House dust atopy and the house dust mite. Leiden: Stafleu's Scientific Publishing Co, 1969:44-52.
5. Cookson JB, Makoni G. Seasonal asthma and the house-dust mite in tropical Africa. Clin Allergy 1975;5:375.

6. Maunsell K, Wraith DG, Cunnington AM. Mites and house dust allergy in bronchial asthma. Lancet 1968;1:1267.
7. Lang JD, Mulla MS. Seasonal dynamics of house dust mites, *Dermatophagoides* ssp., in homes in southern California. Environ Entomol 1978;7:281.
8. Murray AB, Ferguson AC, Morrison B. The seasonal variation of allergic respiratory symptoms induced by house dust mites. Ann Allergy 1980;45:347.
9. Arlian LG, Bernstein IL, Gallagher JS. The prevalence of house dust mites, *Dermatophagoides* spp, and associated environmental conditions in homes in Ohio. J ALLERGY CLIN IMMUNOL 1982;69:527.
10. Wharton GW. House dust mites. J Med Entomol 1976;12:557.
11. Tovey ER, Chapman MD, Platts-Mills TAE. Mite faeces are a major source of house dust allergens. Nature 1981;289:592.
12. Tovey ER, Chapman MD, Wells CD, Platts-Mills TAE. The distribution of dust mite allergen in the houses of patients with asthma. Am Rev Respir Dis 1981;124:630.
13. Tovey ER, Vandenberg R. Mite allergen content in commercial extracts and bed dust determined by radioallergosorbent tests. Clin Allergy 1979;9:253.
14. Woods RA, Eggleston PA, Ingemann L, Schwartz B, Wheeler B, Adkinson Jr NF. Allergen content of household dust samples. J ALLERGY CLIN IMMUNOL 1986;77:130.
15. McCullough JA, Ownby DR. Do commercial house dust extracts (CHDE) contain unique allergens [Abstract]? J ALLERGY CLIN IMMUNOL 1986;77:130.
16. van Bronswijk JEMH, Sinha RN. Pyroglyphid mites (Acari) and house dust allergy: a review. J ALLERGY 1971;47:31.
17. Murray AB, Zuk P. The seasonal variation in a population of house dust mites in a North American city. J ALLERGY CLIN IMMUNOL 1979;64:266.
18. Chapman MD, Platts-Mills TAE. Purification and characterization of the major allergen from *Dermatophagoides pteronyssinus* antigen P_1. J Immunol 1980;125:587.
19. Le Mao J, Dandeau JP, Rabillon J, Lux M, David B. Comparison of antigenic and allergenic composition of two partially purified extracts from *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus* mite cultures. J ALLERGY CLIN IMMUNOL 1983;71:588.
20. Heymann PW, Chapman MD, Platts-Mills TAE. Antigen Der. F_1 from the dust mite *Dermatophagoides farinae:* structural comparison with antigen Der. P_1 from *D. pteronyssinus* and epitope specificity of murine IgG and human IgE antibodics. J Immunol 1986;137:2841.
21. Platts-Mills TAE, Heymann PW, Chapman MD, Hayden ML, Wilkins SR. Cross-reacting and species-specific determinants on a major allergen from *Dermatophagoides pteronyssinus* and *D. farinae:* development of a radioimmunoassay for antigen P_1 equivalent in house dust and dust mite extracts. J ALLERGY CLIN IMMUNOL 1986;78:398.
22. Chapman MD, Heymann PW, Wilkins SR, Brown MJ, Platts-Mills TAE. Monoclonal immunoassays for major dust mite *(Dermatophagoides)* allergens, *Der p* 1 and *Der f* 1, and quantitative analysis of the allergen content of mite and house dust extracts [in press]. J ALLERGY CLIN IMMUNOL.
23. Smith TF, Kelly LB, Heymann PW, Wilkins SR, Platts-Mills TAE. Natural exposure and serum antibodies to house dust mite in mite-allergic children with asthma in Atlanta. J ALLERGY CLIN IMMUNOL 1985;76:782.
24. Hughes AM. The mites of stored food and houses. London: Her Majesty's Stationery Office. 1976.
25. Ford AW, Rawle FC, Lind P, Spieksma FThM, Lowenstein H, Platts-Mills TAE. Standardization of *Dermatophagoides pteronyssinus:* assessment of potency and allergen content in ten coded extracts. Int Arch Allergy Appl Immunol 1985;76:58.
26. Ford A, Seagroatt V, Platts-Mills TAE, and Lowenstein H. A collaborative study on the first international standard of *Dermatophagoides pteronyssinus* (house dust mite) extract. J ALLERGY CLIN IMMUNOL 1985;75:676.
27. Korsgaard J. Mite asthma and residency: a case control study on the impact of exposure to house dust mites in dwellings. Am Rev Respir Dis 1983;128:231.
28. Arlian LG. Humidity as a factor regulating feeding and water balance of the house dust mites *Dermatophagoides farinae* and *D. pteronyssinus* (Acari: Pyroglyphidae). J Med Entomol 1977;14:484.
29. Korsgaard J. House-dust mites and absolute indoor humidity. Allergy 1983;38:85.
30. Mitchell EB, Wilkins SR, Deighton JM, Platts-Mills TAE. Reduction of house dust mite allergen levels in the home: use of the acaricide, pirimiphos methyl. Clin Allergy 15:235, 1985.
31. Fernandez-Caldas E, Swanson MC, Yunginger JW, Reed CE. Immunochemical demonstration of red oak aeroallergens outside the oak pollination season [Abstract]. J ALLERGY CLIN IMMUNOL 1986;77:197.
32. Aalberse RC, Hoorweg E, Reerink-Brongers EE. Interactions between IgE and house dust extract as studied by the radioallergosorbent test (RAST). Dev Biol Stand 1975;29:197.
33. Beug HJ, Dorn M, Strunz M, Thiergart F, Franz U, Windisch S. Pollen grains in house dust. Z Haut Geschlechtskr 1960;29:267.
34. Marsh DG. Allergens and the genetics allergy. In: Sela M, ed. The Antigens, vol III. New York: Academic Press, 1975:271.
35. Bruce CA, Rosenthal RR, Lichtenstein LM, Norman PS. Diagnostic tests in ragweed-allergic asthma. A comparison of direct skin tests, leukocyte histamine release, and quantitative bronchial challenge. J ALLERGY CLIN IMMUNOL 1974;53:230.
36. Rowntree S, Cogswell JJ, Platts-Mills TAE, Mitchell EB. The development of IgE and IgG antibodies to food and inhalant allergens in children at risk of allergic disease. Arch Dis Child 1985;60:727.
37. Platts-Mills TAE, Mitchell EB, Tovey ER, Chapman MD, Wilkins SR. Airborne allergen exposure, allergen avoidance, and bronchial hyperreactivity. In: Kay AB, Austen KF, Lichtenstein LM, eds. Asthma: pysiology, immunopharmacology, and treatment. London: Academic Press, 1984:297.
38. Platts-Mills TAE, Tovey ER, Mitchell EB, Moszoro H, Nock P, Wilkins SR. Reduction of bronchial hyperreactivity during prolonged allergen avoidance. Lancet 1982;2:675-8.